

Heart Homotransplantation in Puppies

Long Survival Without Immunosuppressive Therapy

By YOSHIO KONDO, M.D., FRANZ GRÄDEL, M.D., AND ADRIAN KANTROWITZ, M.D.

THE HEART offers optimum conditions for studying the homograft rejection phenomenon because its histological structure is relatively simple, functioning of the transplant

can be followed precisely, and failure of the transplant immediately causes death of the host. A technique for orthotopic homotransplantation of the heart was developed by Lower and Shumway,¹ who reported successful experiments in 1960. Technical difficulties during surgery resulted in only a very small percentage of survivors. The present report describes a reliable and effective method of heart transplantation in puppies under profound

From the Departments of Surgery of Maimonides Hospital and the State University of New York, Downstate Medical Center, Brooklyn, New York.

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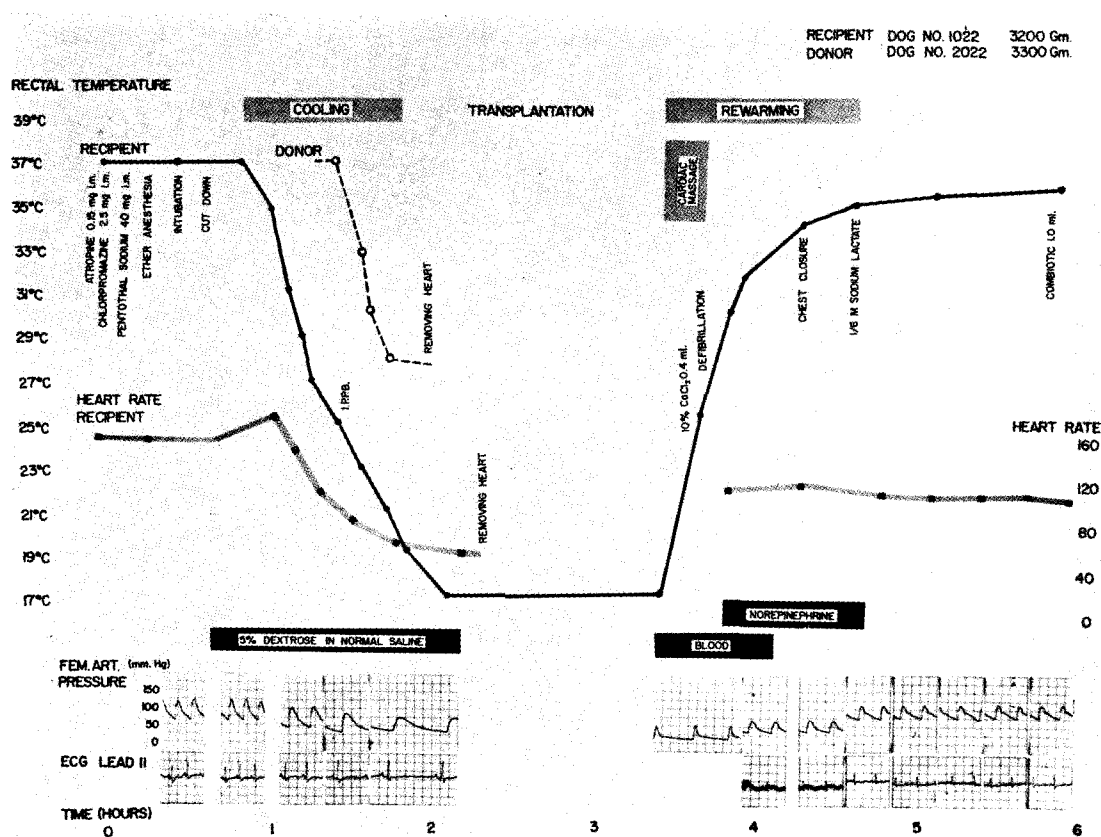


Figure 1

Recipient's rectal temperature is indicated by the solid line and donor's by the dotted line. Heart rate before and after grafting is indicated by the shaded line. Femoral arterial pressure and the electrocardiogram are shown below. Relationship of various steps in the procedure is shown by the time scale. I.P.P.B. = intermittent positive pressure breathing.

Table 1

Survival Data According to Depth of Hypothermia and Type of Graft Preservation in Forty Experiments

EXPERIMENT			RESULT													
NUMBER OF DOGS	TREATMENT OF DONOR	PRESERVATION OF GRAFT	SURVIVAL (DAYS)													
			0-1	-7	-14	-21	-28	-42	-56	-70	-84	-96	-112	-126		
14 (#1001 -1014)	PROFOUND HYPOTHERMIA (7)	NORMAL SALINE	● ●	●												
	NORMOTHERMIA (2)		● ●	●		●										
	MODERATE HYPOTHERMIA (5)		● ●													
26 (#1015 -1040)	MODERATE HYPOTHERMIA	TYRODE SOLUTION	●	● ●	●	● ●	●									
			●	● ●	●	● ●	●			●						
			●	● ●	●	● ●	●							●		

● DIED

○ ALIVE AND WELL

hypothermia without a pump-oxygenator, with survival of more than three months.

Methods

Unrelated pairs of puppies of approximately the same weight, between 3,000 and 4,800 g, were premedicated with atropine sulfate, chlorpromazine, and thiopental sodium and deeply anesthetized with ether. The femoral arterial pressure and electrocardiogram were monitored throughout the procedure. Total body cooling was achieved by ice water immersion. Reduction of the recipient's rectal temperature to 16 to 17 C and the donor's to 27 to 29 C yielded the best results.

The heart was removed and reimplanted by the technique of Lower and Shumway, with a few modifications. The donor heart was kept in Tyrode solution (4 C) for 10 to 30 minutes and sutured in directly opposite fashion. We started at the superior aspect of the right atrium, continued around this chamber and through the atrial septum, then sutured the left atrium, finishing at its inferior part. The aorta and pulmonary artery were then anastomosed. Surgery was performed under complete circulatory arrest, a total of 45 ± 5 minutes. Heart massage was started immediately after replacement. The animal was rewarmed by body immersion and flushing of the chest cavity with warm saline, raising the rectal temperature to 26 to 28 C. With countershock, applied about this time, ventricular fibrillation was readily supplanted by rhythmic contractions. Rewarming was continued until the rectal temperature reached 35 to 36 C. Factors and time relationships of the procedure appear in figure 1.

The animal was kept on liquids through the first postoperative day, and thereafter resumed a regular diet. No attempt was made to suppress the immune reaction. Electrocardiograms, blood samples, and chest X-rays were taken at intervals, and postmortem histological examinations were done on all animals.

Results

Survival Time

Of 40 animals in our series, 24 lived more than one day, and of this number 13 passed the seven-day mark (table 1). One is alive and well on the 112th day. Sixteen puppies, of which the majority were early cases, died within 24 hours, although effective circulation had been restored for more than eight hours in all but three. Causes of death in this group were: ineffective circulation due to early graft failure, brain damage from prolonged circulatory arrest, and other technical failures such as intractable hemorrhage, air embolism, and faultily applied hypothermia. Circulation was maintained adequately in the immediate postoperative period by transplants in all of the last 26 puppies, of which 20 lived more than 24 hours (table 1). Experience as well as improved technique undoubtedly contributed to the higher survival rate.

Postoperative Course

The animals were almost completely conscious within 12 hours. Serum electrolyte bal-

ance was not significantly altered. On the first postoperative day, the puppies walked around and took fluids well. Chest complications such as pneumonia, atelectasis, and empyema—the most annoying problem in canine chest surgery—sometimes resulted fatally within three weeks. Body weight increased steadily, and X-rays showed proportional growth of the heart. The 112-day survivor has moderate

anemia, hypoproteinemia, and lymphocytosis, but is normally active in all respects.

Electrocardiogram and Pulse Rate

In ideal cases, the electrocardiogram and pulse rate indicated no cardiac irregularities. Often, however, the electrocardiogram showed low voltage and various irregularities shortly after resuscitation. These changes usually disappeared on the day after surgery, and there-

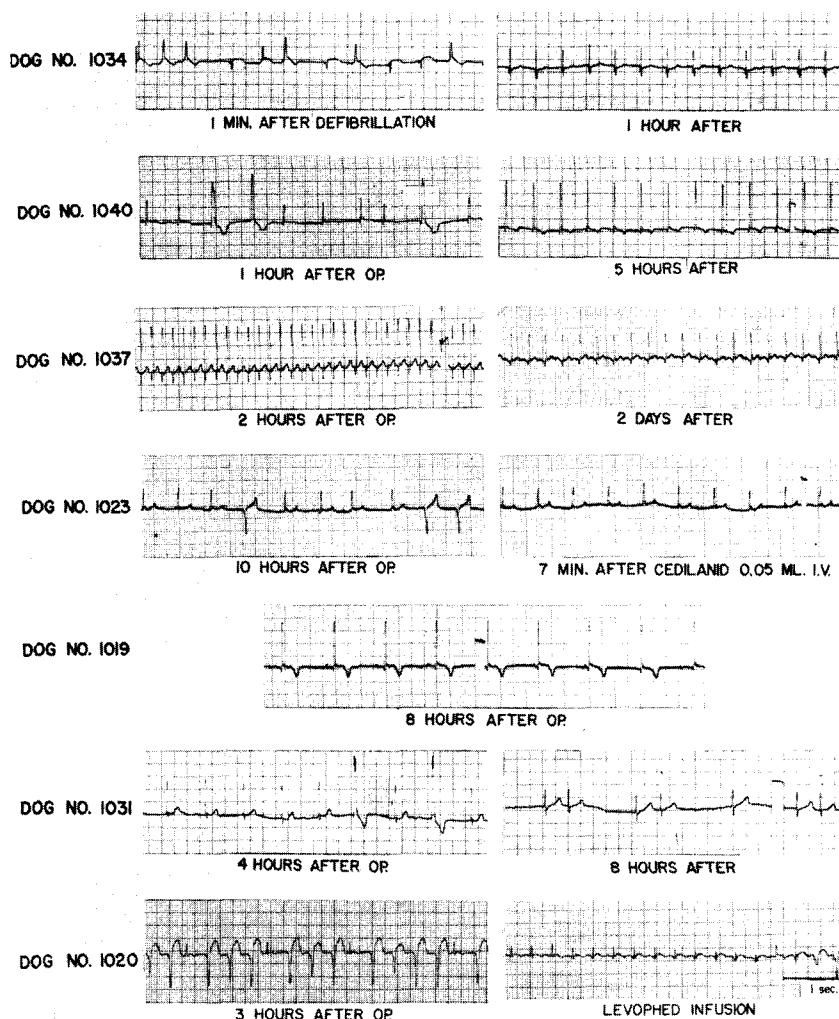


Figure 2

Lead II of irregular electrocardiograms shortly after surgery. Extrasystole, noted after defibrillation in dogs 1034 and 1040, disappeared one to five hours later. Ventricular tachycardia in dog 1037 was not evident 48 hours later. Extrasystole in dog 1023 disappeared soon after intravenous Cedilanid injection, although atrioventricular dissociation persisted. This common finding early in the postoperative period is also shown in the electrocardiogram of dog 1019. Electrocardiograms of dogs 1031 and 1020 are typical in cases of graft failure.

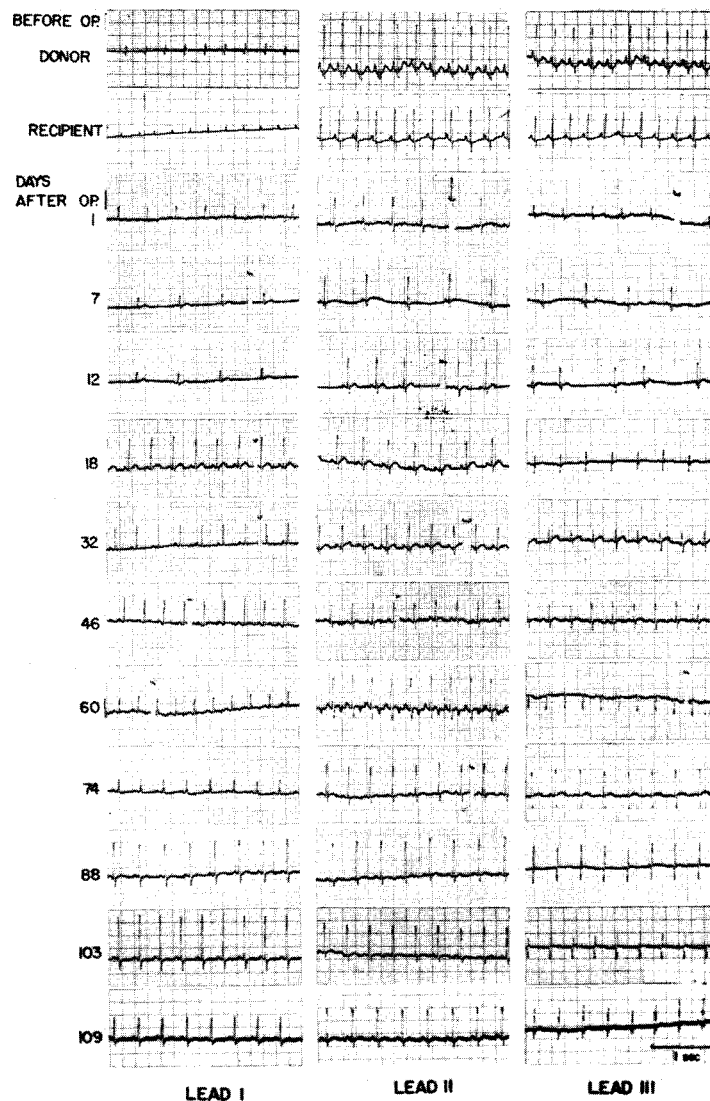


Figure 3

Electrocardiograms of donor and of recipient (dog 1022), taken pre-operatively and at intervals ranging from 1 to 109 days postoperatively.

after the pulse was fairly regular, with a constant rate of 110 to 150/min (fig. 2). Follow-up electrocardiograms revealed the activity of the transplant (fig. 3). Also, the final fate of the transplant could be predicted in several instances from such signs as low voltage and irregular rhythm before the animal's general condition had begun to deteriorate.

Histopathology

The following microscopic findings were

typical of the series. Dog no. 1018 (57-day survivor) died of congestive heart failure. The autopsy revealed a slightly enlarged heart and a greatly enlarged liver with 1,000 ml of ascitic fluid. The anterior wall of the heart showed a recent myocardial infarct and focal necrosis of the papillary muscle (fig. 4). Among other findings (fig. 5) were: acute endocarditis and thromboangiitis of the left circumflex artery, chronic passive congestion

of the liver, and passive congestion of the spleen without follicular hyperplasia.

In dog no. 1016 (15-day survivor), the heart revealed endocarditis, focal myocarditis, and epicarditis with focal calcific deposits. The spleen showed moderate reticular cell hyperplasia.

In dog no. 1029 (12-day survivor), fairly extensive myocarditis was observed in all heart sections. The myocardial section crossing the suture line indicated that this condition was more severe in the graft than in the host.

Virtually normal myocardial tissue was noted in dog no. 1003 (5-day survivor), dog no. 1011 (20-day), and dog no. 1023 (25-day).

Discussion

In the many reports of heterotopic homotransplantation,^{2,3} with implantation of a

puppy heart in the neck or groin of an adult dog imposing no load on the left ventricle, survival rarely exceeded ten days. Lower and Shumway^{1,4} reported a 21-day survival following orthotopic homotransplantation of the heart in dogs without immunosuppressive treatment. Blumenstock et al.⁵ performed 50 such experiments, instituting Methotrexate treatment after surgery. Eight of the 50 dogs lived more than one day, and one lived 42 days.

A much higher percentage of the animals in our series lived more than one day, and the rejection process was manifest much later. One puppy is alive and well on the 112th day. Although this success may be due to accidental histocompatibility, such cases—together with successful heart autotransplant studies^{6,7}—suggest an encouraging future for heart transplantation.

We have started to use immunosuppressive

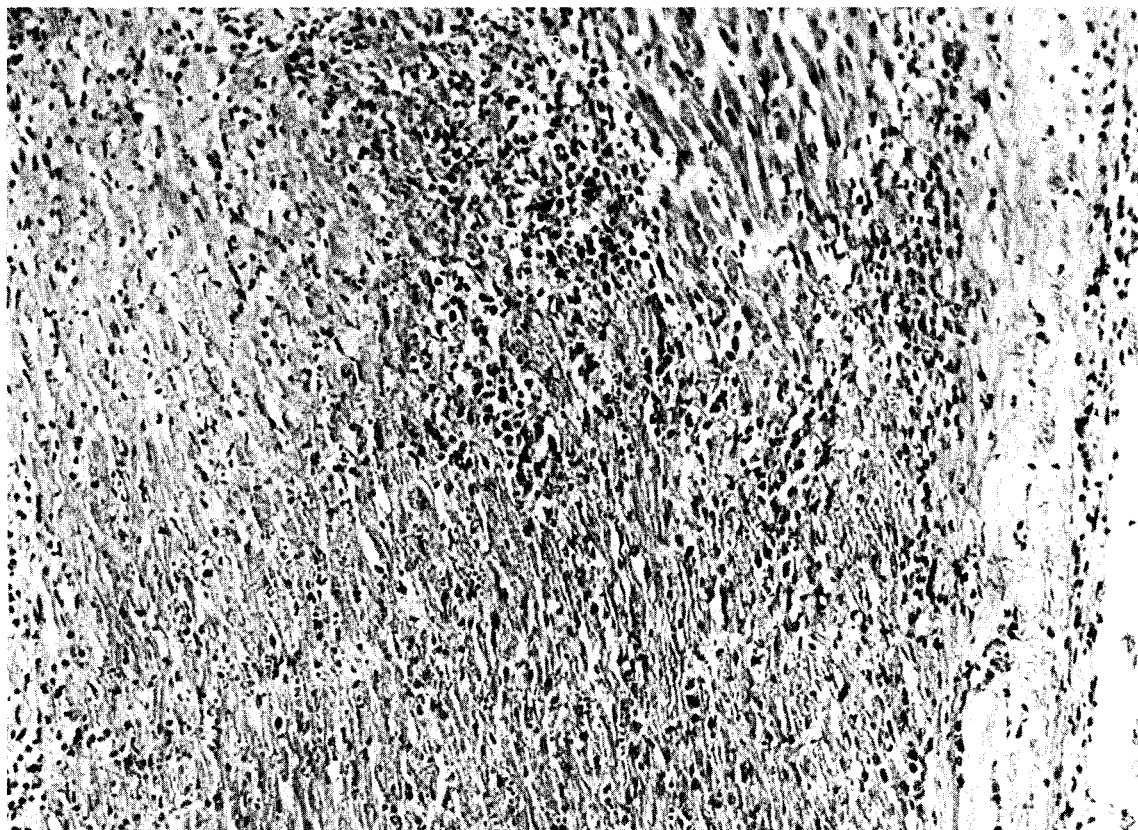
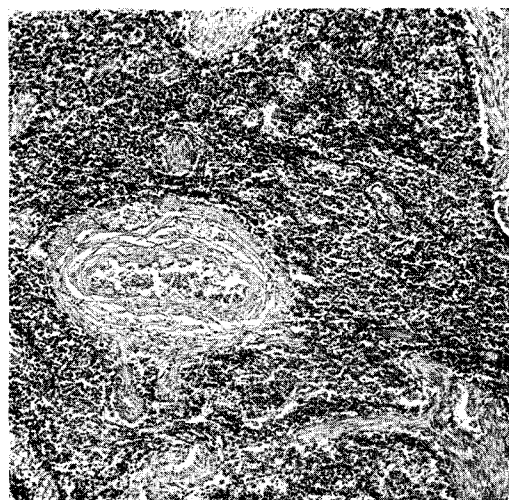
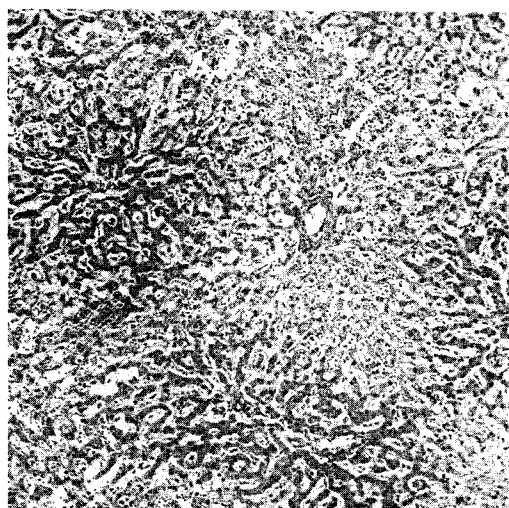
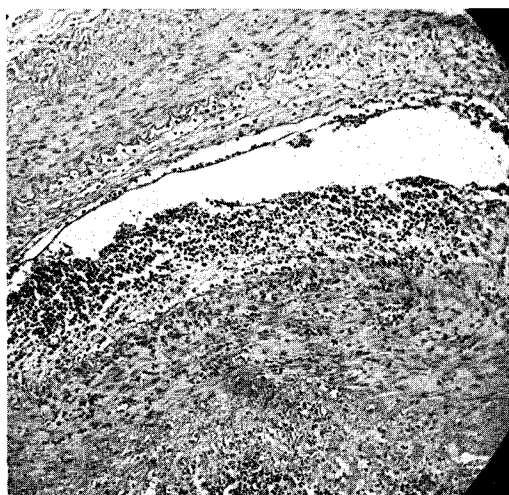


Figure 4

Focal necrosis of the papillary muscle in 57-day survivor. Hematoxylin and eosin, $\times 400$.



treatment (Imuran) in a few cases. Also, we have transplanted heart grafts preserved 24 hours; one such puppy is alive and well on the fifth postoperative day. Data will be reported in the near future.

Summary

A satisfactory technique for orthotopic homotransplantation of the heart, under profound hypothermia without a pump-oxygenator, has been presented. Of the 40 puppies in our series, 24 lived more than one day; 13 lived more than seven days. One is well and active on the 112th day. The rhythm of the transplanted hearts was often somewhat irregular shortly after surgery, but usually became fairly regular on the following day and maintained a constant rate even after exercise. Long survivors steadily gained weight and showed a proportional increase in heart size. The fate of the transplant could be forecast in some instances from the follow-up electrocardiograms. Although the histological findings were not uniform, acceptance of the homologous heart greatly exceeded our expectations. Unpredictable histocompatibility may possibly explain the long survivors.

Acknowledgment

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Figure 5

Other autopsy findings in 57-day survivor include thromboangiitis of the coronary artery (top); chronic passive congestion of the liver (middle); and passive congestion of the spleen (bottom). Hematoxylin and eosin, $\times 210$.

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